

## PATENT COOPERATION TREATY

## PCT

## INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 8799/9	<b>FOR FURTHER ACTION</b> See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)	
International application No. PCT/US98/19494	International filing date (day/month/year) 18/09/1998	Priority date (day/month/year) 19/09/1997
International Patent Classification (IPC) or national classification and IPC C12N15/53		
Applicant PROMEGA CORPORATION et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.


2. This REPORT consists of a total of 8 sheets, including this cover sheet.

- ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

These annexes consist of a total of 4 sheets.

3. This report contains indications relating to the following items:

- I ☒ Basis of the report
- II ☐ Priority
- III ☒ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- IV ☐ Lack of unity of invention
- V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- VI ☒ Certain documents cited
- VII ☐ Certain defects in the international application
- VIII ☒ Certain observations on the international application

Date of submission of the demand  16/04/1999	Date of completion of this report  11.01.00
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**I. Basis of the report**

1. This report has been drawn on the basis of (*substitute sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to the report since they do not contain amendments.*):

**Description, pages:**

1-76 as originally filed

**Claims, No.:**

1-35 with telefax of 09/12/1999

**Drawings, sheets:**

1/59-59/59 as originally filed

2. The amendments have resulted in the cancellation of:

- ☐ the description, pages:  
☐ the claims, Nos.:  
☐ the drawings, sheets:

3. ☒ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)):

**see separate sheet**

4. Additional observations, if necessary:

**III. Non-establishment of opinion with regard to novelty, inventive step and industrial applicability**

The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non-obvious), or to be industrially applicable have not been examined in respect of:

- ☐ the entire international application.  
☒ claims Nos. 2, 4, 7, 8, 10, 13-15, 18, 22, 35.

because:

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- ☒ the said international application, or the said claims Nos. 2, 4, 7, 8, 10, 13 relate to the following subject matter which does not require an international preliminary examination (*specify*):

**see separate sheet**

- ☒ the description, claims or drawings (*indicate particular elements below*) or said claims Nos. 22 are so unclear that no meaningful opinion could be formed (*specify*):

**see separate sheet**

- ☐ the claims, or said claims Nos. are so inadequately supported by the description that no meaningful opinion could be formed.

- ☒ no international search report has been established for the said claims Nos. 14, 15, 18, 35.

## V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

### 1. Statement

Novelty (N)	Yes:	Claims	1, 3, 5, 6, 9, 11, 12, 16, 17, 19-21, 23-34
	No:	Claims	
Inventive step (IS)	Yes:	Claims	1, 3, 5, 6, 9, 11, 12, 16, 17, 19-21, 23-34
	No:	Claims	
Industrial applicability (IA)	Yes:	Claims	1, 3, 5, 6, 9, 11, 12, 16, 17, 19-21, 23-34
	No:	Claims	

### 2. Citations and explanations

**see separate sheet**

## VI. Certain documents cited

### 1. Certain published documents (Rule 70.10)

and / or

### 2. Non-written disclosures (Rule 70.9)

**see separate sheet**

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**VIII. Certain observations on the international application**

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

**see separate sheet**

### **Citations**

The documents mentioned in this International Preliminary Examination Report (IPER) are numbered as in the International Search Report dated 19.03.99, i.e. **D1** and **D6** correspond to the first and the last document of the search report, respectively.

### **Re ITEM I (Basis of the opinion)**

- 1 The amended claims 1, 3, 5, 6, 9, 11, 12 and 14-35 filed with telefax of 09.12.99 can be regarded as meeting the requirements of Art. 34(2)(b) PCT.
- 2 The amended claims 2, 4, 7, 8, 10 and 13 filed with telefax of 09.12.99 contain subject-matter which extends beyond the disclosure in the international application as filed and therefore contravene Art. 19(2) PCT. The following amendments are not allowable:
  - 2.1 Claims 2 and 4: no basis could be found for the features "five/ten hours in an aqueous solution at about 50°C". The expression "at about 50°C" renders the scope of protection broader than "at 50°C" (cf. description p. 4, l. 13-14 and original claims 4 and 8).
  - 2.2 Claims 7 and 8: no basis could be found for the features "six hours/two days in an aqueous solution at about 60°C". The expression "at about 60°C" renders the scope of protection broader than "at 60°C" (cf. p. 4, l. 22 and p. 4, l. 24-25).
  - 2.3 No basis can be found for the subject-matter of claim 10 ("at least 95% of its activity after six weeks at 22°C") (cf. p. 4, l. 4: "loss in activity of 5%", i.e. there is only a basis for "less than" or "up to" 95% of its activity. The expression "at least 95%" means 95% or more).
  - 2.4 No basis can be found for the subject-matter of claim 13 ("wherein each mutation changes the native amino acid to a consensus amino acid").

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According to Rule 70.2(c) PCT, this IPER has been established as if some of the amendments had not been made, since they have been considered to go beyond the disclosure as filed. Therefore, under Rule 70.2(c) PCT (also see **ITEM III**) examination is being carried out on the basis of claims 1, 3, 5, 6, 9, 11, 12 and 14-35 filed with telefax of 09.12.99 but not for the subject-matter of amended claims 2, 4, 7, 8, 10 and 13 for which no basis could be found in the original claims, description or drawings.

**Re ITEM III (Non-establishment of opinion)**

- 1 No search report has been established for the subject-matter of original claims 16, 21 and 22. Thus, preliminary examination has not been carried out for the sequences defined in amended claims 14, 15, 18 and 35; said claims correspond to original claims 16, 21 and 22 (Rule 66.1(e) PCT).
- 2 For the reasons stated under **ITEM I, 2**, no examination has been carried out for the subject-matter of amended claims 2, 4, 7, 8, 10 and 13.
- 3 Claim 22 has not been examined because no use is indicated, i.e. the essential technical feature is missing (Art. 6 PCT). Claims 23-29 have been examined as if they were directed to uses of a synthetic luciferase according to any of claims 1-15.
- 4 In summary, examination has been restricted to the subject-matter of amended claims 1, 3, 5, 6, 9, 11, 12 and 31-34 (completely) and amended claims 16, 17, 19-21 and 23-30 (partially) in so far as they refer to the subject-matter of claims 1, 3, 5, 6, 9, 11 and 12.

**Re ITEM V (Novelty, inventive step, industrial applicability)**

**1 Summary of the present application**

The present application is related to thermostable beetle luciferases and a process for the identification of thermostable beetle luciferases.

**2 Novelty (Art. 33(2) PCT)**

- 2.1 The subject-matter of claims 1, 3, 5, 6, 9, 11, 12, 16, 17, 19-21 and 23-34 has not been made available to the public by any of the available prior art documents and can therefore be regarded as novel.

**D1** discloses a beetle (firefly) luciferase which remains 65% or more of its activity after 60 min at 50°C (D1, p. 4, l. 30-31). **D2** discloses a beetle (firefly) luciferase with a half life of 30 min at 43.5°C in 50mM potassium phosphate buffer pH 7.8 (D2, p. 12, Example 2). **D3** discloses a beetle (firefly) luciferase with a half life of 12 min at 40°C (see Table 1 of D3). However, said documents do not disclose a beetle luciferase with a half life of two hours at about 50°C or a polynucleotide which encodes for such a luciferase (independent claims 1 and 16) (attention is drawn to **ITEM VIII, 1**).

- 2.2 Uses according to claims 23-29, viz. of a synthetic luciferase with a half life of two hours at about 50°C, and the hybrid protein of claim 30 have also not been disclosed in the available prior art documents and can therefore be regarded as novel.
- 2.3 Also the method according to independent claim 31 wherein recursive mutagenesis and selection ("directed evolution", cf. p. 5, l. 29-p. 6, l. 8) is applied to prepare a mutant beetle luciferase with a half life of two hours at about 50°C has not been disclosed in the available prior art.

**3 Inventive step (Art. 33(3) PCT)**

The subject-matter of claims 1, 3, 5, 6, 9, 11, 12, 16, 17, 19-21 and 23-34 cannot be derived from the available prior art in an obvious manner and therefore complies with the requirements of Art. 33(3) PCT.

**4 Industrial applicability (Art. 33(4) PCT)**

Claims 1, 3, 5, 6, 9, 11, 12, 16, 17, 19-21 and 23-34 meet the criteria as set forth by Art. 33(4) PCT.

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**Re ITEM VI (Certain published documents (Rule 70.10))**

Application No Patent No	Publication date (day/month/year)	Filing date (day/month/year)	Priority date (valid claim) (day/month/year)
WO-A-98 46729	22.10.98	07.04.98	11.04.97

Said document has been published after the filing date of the present application and does therefore not constitute prior art in the meaning of Rule 64(1)(b) PCT. However, **D6** claims an earlier priority than the present application and will therefore become of relevance for the novelty of the claimed subject-matter during regional phase examination.

**Re ITEM VIII (Certain observations on the international application)**

- 1 Rule 6.3(a) PCT requires that the matter for which protection is sought be defined in terms of technical features of the invention (also cf. PCT Guidelines III-4.4, as in force from 09.10.98). A peptide (claims 1, 3, 5, 6, 9, 11, 12 and 30) and a polynucleotide (claim 16) are chemical compounds which can be clearly and unambiguously defined by their chemical structure, i.e., their amino/nucleic acid sequence.

The IPEA is of the opinion that the definition of the half life at a certain temperature and time is not a sufficient way to define a protein. As a general rule, the characterization of a chemical compound solely by its parameters is not allowable (cf. PCT Guidelines III-4.7a). Attention is drawn to the fact that none of the specific sequences defined in Figures 22-47 have been searched by the ISA (see also **ITEM III, 1**).